

HeLa-EGFRVIII cells were generated from the human cervical cancer cell line HeLa by transfection with an expression vector encoding EGFRVIII with a C-terminal FLAG tag, followed by selection in medium containing geneticin. Expression of EGFRVIII was confirmed by flow cytometry using cells that were fixed and permeabilized (Figure 1).

<b>Storage</b>
Store vial in liquid nitrogen immediately upon receipt.
<b>Formulation</b>
Cells are cryopreserved in 0.5 ml - 1 ml of 90% FBS + 10% DMSO.
<b>Thaw Protocol</b>
Partially immerse the vial in a 37°C water bath with gentle shaking until most of the medium is thawed. In a tissue culture hood, add 1 ml of pre-warmed culture medium into the vial and immediately transfer the contents of the vial to a centrifuge tube containing 5-10 ml of pre-warmed culture medium. Centrifuge the tube at room temperature for 5 minutes, aspirate the supernatant and suspend the cell pellet in 5-10 ml of pre-warmed culture medium.
<b>Culture Protocol</b>
HeLa-EGFRVIII is an adherent cell line. Culture the cells in DMEM containing 10% FBS and 1 mg/ml geneticin using a humidified incubator set to 5% CO <sub>2</sub> . When the cell monolayer is nearly confluent, use trypsin-EDTA to detach the cells from the culture flask and immediately neutralize the trypsin by adding fresh, pre-warmed culture medium to the cells. Cells should be passaged at least twice per week.
<b>Notices &amp; Disclaimer</b>
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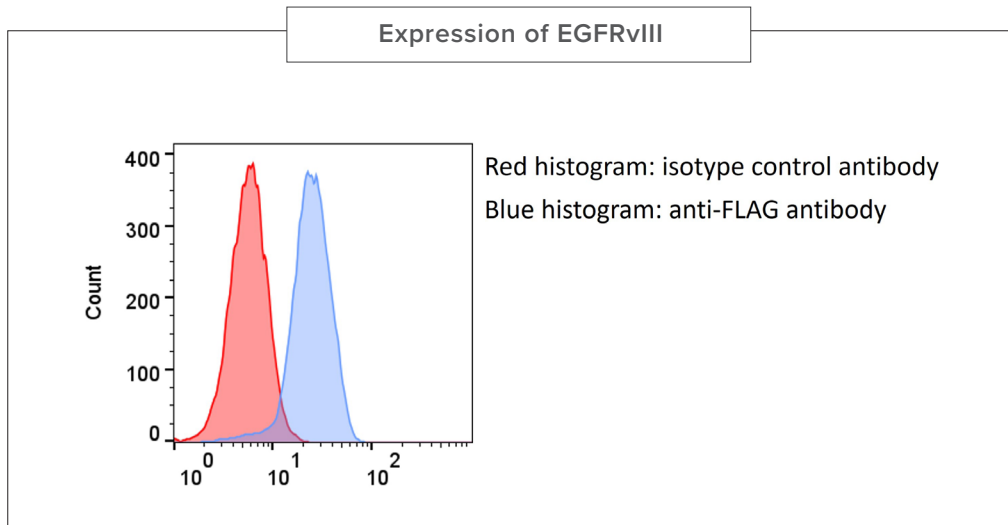


Figure 1. PM-EGFRVIII-HELA cells were fixed, permeabilized and stained with an anti-FLAG antibody or an isotype control antibody. The FLAG antibody bound to over 95% of the PM-EGFRVIII-HELA cells.

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